

Project Title

Characterisation of neuropathic pain in children: multimodal assessment and diagnosis.

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1. Introduction

Can patient-reported outcomes, sensory testing, and brain imaging improve the diagnosis and understanding of neuropathic pain in children?

In some children with chronic pain, pain is related to damage or diseases that affect the nerves that send pain signals. This is known as neuropathic pain. This is not well understood and can be difficult to diagnose. It can often produce unusual feelings such as sensitivity of the skin to light touch. Neuropathic pain is often severe and difficult to treat, and can affect quality of life for the child and family. In order to improve management, it is important to improve recognition of this condition, and understand what factors and mechanisms are associated with neuropathic pain in children.

The primary aim of this project is to determine whether a questionnaire to identify neuropathic pain in adults is also useful in children. This will help paediatricians and other doctors to recognise neuropathic pain and start appropriate treatment or know when to refer children to a chronic pain clinic. We will also measure the effects of neuropathic pain on mood, sleep, and quality of life (using questionnaires); identify changes in the sensitivity of the skin to touch and other sensations (using specialised sensory tests); and determine the feasibility of brain imaging for assessing changes in the brain (using magnetic resonance imaging or MRI), in these children. These secondary aims are exploratory and will enable the design of further studies that will allow us to measure how effective different types of treatment are, and help ensure that children get the most appropriate treatment or interventions to reduce pain and effects on quality of life.

10-18 year old children diagnosed with chronic pain arising from neuropathic and non-neuropathic origins will be recruited from the Chronic Pain Outpatients service at Great Ormond Street Hospital NHS Trust. Results from this pilot study will inform future studies.

2. Aims of study

This study will recruit children and adolescents aged 10-18 years referred to the Great Ormond Street Hospital Pain Service who are clinically diagnosed as having chronic pain from neuropathic or non-neuropathic origin. Our overall aim is to:

1. develop better **outcome tools** to screen for neuropathic pain and assess the impact of chronic neuropathic pain in children;
2. comprehensively characterise the **symptoms and signs of neuropathic pain** related to **different causes and mechanisms**;
3. evaluate the utility and feasibility of **specialized assessment** of pain processing with quantitative sensory testing (QST) and **neuroimaging** to elucidate neuropathic pain mechanisms.

3. Objectives

The principal objective is to assess the clinical utility of a neuropathic pain screening tool against the gold standard of clinical diagnosis, consistent with adult validation studies.

Secondary objectives are exploratory and will help design further studies. These are:

1. To characterise the effects of neuropathic pain on quality of life, mood, and function by comparing to healthy controls and children with other types of chronic pain. This will help us to understand whether some factors are particularly associated with neuropathic pain, and could help to improve treatment.
2. To understand the pattern of altered sensitivity to touch and other sensations in children with neuropathic pain. This will inform us about the function of the different nerve fibres that carry information about touch and pain. This will help understand the mechanisms/causes associated with neuropathic pain.
3. To assess a) the feasibility of brain scanning and of using sensory stimuli during scanning; and b) the acceptability of brain scanning, in children with neuropathic pain.
4. To carry out preliminary analysis of brain changes associated with neuropathic pain in children.

Secondary objectives 1 & 2 are based on evaluation of data currently collected as part of routine clinical practice by the GOSH Pain Service. Objective 1: Researchers will access questionnaire data following review by the multidisciplinary team (pain physician/doctor, clinical psychologist, clinical nurse specialist, physiotherapist) at Chronic Pain Outpatients. Objective 2: Quantitative Sensory Testing will be performed and the clinical care team will receive a written clinical report to guide further management. The proposed research will not alter routine care.

Secondary objectives 3 & 4 will inform a future more comprehensive brain scanning study. To assess brain changes associated with neuropathic pain we will compare with existing data from children of the same age without neuropathic pain, which has been previously collected by our research collaborators at GOS Institute of Child Health.

4. Hypothesis

4a. Primary Hypothesis

The neuropathic pain screening tool has adequate sensitivity and specificity to identify neuropathic pain in children.

4b. Secondary Hypotheses

1. Scores on questionnaires assessing quality of life, mood, and function are different in children with neuropathic pain compared to healthy controls.

2.
 - a. Children with neuropathic pain have altered sensitivity to touch or other sensations relative to healthy controls;
 - b. Patterns of altered sensitivity in children with neuropathic pain match sensory profiles identified in adults.
3. MRI and fMRI neuroimaging is feasible and acceptable in children with neuropathic pain.
4. There are structural and functional changes in brain regions involved in pain signalling in children with neuropathic pain.

5. Study Design

This is a cross-sectional study utilising questionnaires and specialised sensory testing methods (QST) in 10 – 18 year old children and adolescents with neuropathic pain. In a subset of participants, this will also be a feasibility study for neuroimaging, for a potential future neuroimaging study. This design is appropriate because essential first steps for improving the management of neuropathic pain in children include: i) improving recognition and diagnosis of this type of pain in children (which is our primary aim); ii) development of outcome tools for use in future controlled trials; and iii) to monitor the clinical progression/resolution of pain and treatment response. Our secondary aims involve the use of patient-reported outcomes, QST, and neuroimaging to explore their clinical and research utility as outcome tools.

6. Study Location

This is a single-centre study that will be carried out at an Outpatient clinic (Chronic Pain Service clinic) at the Great Ormond Street Hospital NHS Trust. Sensory testing will be carried out as part of the Outpatient appointment at the Sensory Testing Room, while neuroimaging will be carried out in the Imaging Suite, at Great Ormond Street Hospital NHS Trust.

7. Study Population

The Pain Service at Great Ormond Street Hospital NHS Trust manages a large and unique population of inpatients and outpatients with neuropathic pain (one-third of chronic pain referrals), related to a range of causes that include persistent post-surgical pain, trauma, different medical conditions, and rare genetic diseases. This study will recruit 74 children and adolescents aged 10-18 years referred to the Great Ormond Street Hospital Pain Service who are clinically diagnosed as having neuropathic pain, and 150 who are diagnosed as having chronic pain of non-neuropathic origin. All male and female children within this age range that are referred to the Pain Service will be approached.

8. Eligibility Criteria

8a. Inclusion criteria

Children aged 10-18 years with a clinical diagnosis of chronic pain of either neuropathic or non-neuropathic origin will be included. Children in this age range can describe a range of pain descriptors, complete validated patient-reported outcomes, and both understand and tolerate QST.

8b. Exclusion criteria

Children with significantly impaired comprehension (less than school level for 10 year old) or inadequate English language skills will be excluded as questionnaires are validated in English and sensory testing instructions can only be delivered by the Investigators in English. For brain imaging, patients will be excluded if they have: (i) significant medical illness or other (non-neuropathic) neurological disease; (ii) pregnancy; and (iii) magnetic implants of any type.

9. Study Outcomes

9a. Primary Outcome

The primary outcome is the sensitivity and specificity of a neuropathic pain screening tool in distinguishing between chronic pain patients with pain arising from a neuropathic origin vs. those with pain arising from a non-neuropathic origin: the Self-Report Leeds Assessment of Neuropathic Symptoms and Signs (S-LANSS) questionnaire.

9b. Secondary Outcomes

Secondary outcome measures are exploratory, and will inform a future more comprehensive study about likely effect sizes. These outcome measures will be collected only in children with neuropathic pain, and include:

1. Scores on patient- and parent-reported outcome measures of quality of life, mood, and function. We will compare with existing control data, published healthy control values, and with data from the existing chronic pain database for a preliminary analysis of whether the severity of symptoms and disability are similar to, or greater than, other causes of chronic pain in children.
2. Sensory profiles. QST data for each patient will be compared with existing healthy control data and with published control data to determine their sensory profile in terms of gain or loss of sensitivity across all QST parameters. We will then assess: 1) the proportion of patients having sensory changes for each QST parameter; 2) whether the overall sensory profiles cluster according to the cause of neuropathic pain and according to profiles identified in adult research.
3. Neuroimaging. We will assess the recruitment rate for neuroimaging, and, in some children who assent, the feasibility of using sensory stimuli during scanning, in this cohort. Preliminary analysis will assess grey and white matter integrity of brain regions involved in pain signalling relative to existing control data, as well as changes in functional connectivity between these key brain regions. If stimulation is feasible, we will also compare which brain areas are activated, and to what degree, following stimulation of the painful and non-painful body area in children with neuropathic pain.

10. Study Procedures

10a. Recruitment of participants

Children will be recruited from the Chronic Pain Outpatients Service.

Children in the study will be aged between 10 and 18 years, and information will be given in an age-appropriate manner (separate information sheets for 10-15 and 16-18 year olds). If consent and assent are obtained, the participant will be assigned a unique Study Number which will be used in all records. Copies of clinical assessments to be included in the study (questionnaire results) will have personal identification data removed and be retained only with the study number.

We will recruit 74 children with neuropathic pain and 150 children with non-neuropathic chronic pain within a period of 3 years. This sample size is based primarily on a power calculation to identify a suitable sample size for the primary objective, and is sufficient for the secondary objectives as well. Details are provided in Section 11a.

10b. Randomisation

No intervention is being given, therefore randomisation is not required for this study.

10c. Study procedure

Testing will only be carried out in children and parents who have given informed consent/assent. Children with non-neuropathic chronic pain will only need to complete a neuropathic pain screening tool (given routinely but repeated for research purposes). Children with neuropathic pain and their parents will be asked to complete questionnaires and QST, and if they wish, a separate visit for a brain scan will be arranged, as outlined below.

Questionnaires will include: a pain history to measure pain levels, and a neuropathic pain screening tool (given routinely but repeated for research purposes at the time of testing). If routine questionnaires (described in Section 10d) assessing quality of life, anxiety, depression, sleep disturbance, and pain coping have not been administered within the last 3 months during a clinic appointment, they will be repeated at this time.

Next, QST will be performed. This is a standard procedure, with patients attending the Pain Service clinic increasingly being referred for QST, and here we aim to formalize assessment in all patients with neuropathic pain. As detailed in Section 10d, established guidelines exist for use of QST in clinical trials and in assessment of neuropathic pain. QST consists of a series of standardised sensory tests, used previously to measure sensory function in normal children and children with chronic pain. QST will be performed using established standardised protocols and verbal instructions, in the Pain Service sensory testing room, GOSH NHS Trust. With their parent present, the child will be told about each test and given a short demonstration. Testing will be performed on the palm of the non-dominant hand at the base of the thumb to evaluate generalized changes in sensitivity, and allow comparison with existing control reference data. Testing will also be performed near the site of pain or previous tissue injury/scars, and where feasible (i.e. unilateral symptoms, such as in CRPS), on the opposite side of the body for comparison, as QST measures vary across different body sites.

If parents and children consent/assent to a brain scan, a separate visit will be arranged. Patients and their family will be asked to fill in and check the MRI safety questionnaire with the radiographer. If participants are willing to consider additional evaluation, they will receive sensory stimulation within the scanner in addition to the resting state scans. Testing will be performed on a body site with no pain and then near the site of pain or previous injury/scars. Stimuli will include the MRI-compatible brush, thin nylon filament, and blunt metal probe that are used in QST. During scanning, each stimulus will be applied up to 5 times at each body site, and patients will score how uncomfortable this is.

Following scanning, patients and their parents/carers will be asked about their experience with the scanning procedure using a standardised questionnaire.

10d. Measurement tools used

Patient- and Parent-reported Outcomes: domains and assessment tools

We aim to quantify quality of life, sleep, mood and function in children with neuropathic pain, and compare these outcomes with sensory and brain changes. These patient-reported outcome measures are routinely collected during initial assessment of new patients by the Chronic Pain Service, and at subsequent follow up outpatient appointments to monitor progress. These include:

1. Paediatric Quality of Life Inventory (PedsQL, Child and Parent Report). This tool evaluates 23 items relating to the child's functioning in four dimensions: physical; emotional; social; and school. The Parent Report assesses parents/carers' perceptions of their child's quality of life. Age-appropriate scales are used.
2. Pain Catastrophizing Scale (Child [PCS-C] and Parent [PCS-P] versions). Catastrophizing about pain is characterized by 13 items assessing 3 aspects of pain experience: the tendency to magnify the threat value of pain (magnification), to feel helpless in the context of pain

(helplessness), and to worry about it more (rumination). The PCS-C is validated for children aged 9-17 years, while the parent version (PCS-P) relates to the thoughts and feelings of the parent/carer when their child is in pain. The scale has a maximum score of 52, with low scores (0-14) reported by healthy control groups, scores of 15-25 ranked as moderate, and ≥ 26 high. Scores over 20 have been reported in chronic pain populations.

3. Pain Coping Questionnaire (PCQ). The PCQ comprises 39 items relating to information seeking; problem solving; seeking social support; positive self-statements; behavioural distraction; cognitive distraction; externalising; and internalising/catastrophizing. These subscales load onto 3 scales: approach; problem-focused avoidance; and emotion-focused avoidance.
4. Adolescent Sleep-Wake Scale-Revised (ASWS-R). The ASWS-R evaluates 10 items relating to: falling asleep and reinitiating sleep; returning to wakefulness; and going to bed.
5. Paediatric Index of Emotional Distress (PI-ED). The PI-ED comprises 14 items related to symptoms of anxiety and depression, with a maximum score of 42 and a threshold of 20 indicating risk of developing comorbid anxiety and depression.
6. Hospital Anxiety and Depression Scale (HADS). The HADS measures anxiety and depression in adults and comprises 14 items. Scores range from 0 to 21 for each scale, with suggested cut-off scores of 0-7 indicating normal anxiety/depression; 8-10 mild; 11-14 moderate; and 15-21 severe anxiety/depression.

Neuropathic pain screening tools

Recognition and early identification of neuropathic pain is important for providing effective analgesia and evaluating potential preventative or protective strategies. Several screening tools for neuropathic pain are used in adults. The S-LANSS has demonstrated 74% sensitivity and 76% specificity as a screening tool for neuropathic pain in adult chronic pain populations, and includes 5 questions related to symptoms and pain descriptors and 2 related to examination (response to rubbing or pressing in painful vs. non-painful area). In adults, scores $>12/24$ are indicative of neuropathic pain. An important aim of this project is to **compare scores on the S-LANSS** with the **clinical diagnosis** and **QST results** to assess its clinical utility as a screening tool in children with neuropathic pain. The S-LANSS is currently routinely administered during the Outpatients appointment but will be repeated at the time of the study for all patients to facilitate comparison with sensory testing.

Pain history

We will ask children and their parent(s) information about what type of pain they feel, its duration, frequency, and location, aggravating and relieving factors, and what pharmacological and non-pharmacological management they have received in the past. We have designed a standardised questionnaire to capture this information.

We will also ask children about the intensity of their pain using visual analog scales to assess pain intensity on three items: current pain, average pain over the last week, and worst pain over the last week. On a 10 cm line with anchors of 0 and 10, the participant marks the point representing the intensity of their pain.

Quantitative Sensory Testing (QST)

Quantitative Sensory Testing (QST) incorporates a range of somatosensory stimulus modalities and intensities to evaluate A-beta large myelinated fibre function, as well as small unmyelinated C and myelinated A-delta fibre function that is relevant for pain processing but not evaluated with clinical nerve conduction studies. QST provides a comprehensive picture of localized and generalized decreases and/or increases in sensitivity. Conditioned Pain Modulation (CPM) protocols assess endogenous pain modulation, mediated by different brain regions converging on brainstem pain control centres to send descending pathways to the spinal cord and facilitate or inhibit pain

processing. A noxious 'conditioning stimulus' alters sensitivity to a 'test stimulus' at a distant body site, with increased inhibition reflected by an increased threshold (variable test stimulus).

Specific guidelines have been established for use of QST in clinical trials in both adults and children, and for the evaluation of neuropathic pain related to a range of causes in adults. Standardised protocols and verbal instructions will be used in the current study to assess the following modalities:

Static thresholds

Thermal detection thresholds. Participants press a response button on detection of cool (CDT) or warm (WDT), or when discomfort is experienced by cold (cold pain threshold, CPT) or heat (HPT). Each stimulus is repeated 5 times, and the threshold taken as the mean. Subjects are also asked to report rapid changes in temperature or paradoxical sensations (hot or burning sensations felt as cold, or cold perceived as hot).

Mechanical detection threshold (MDT). Hand-held von Frey monofilaments with logarithmically increasing bending force are applied to the skin at right angles with sufficient force to bow the filament. With eyes closed, participants report "yes" when they feel light touch. Appearance and disappearance thresholds are sequentially determined, using an up-down method, with the threshold calculated as the geometric mean of ten values.

Mechanical pinprick threshold (MPT). From an ascending sequence of weighted pinprick stimulators (8, 16, 32, 64, 128, 256 and 512 mN) (Rolke et al., 2006), participants report the stimulus that produces an uncomfortable pricking sensation rated on a 0-10 verbal rating scale (0=not pricking; 10=intense pricking as produced by a needle/blood test).

Pressure pain threshold (PPT). A computer controlled hand-held 1cm² algometer (Somedic SENSEBox) incorporating an optical feed-back system is used to ensure a standardised increase in pressure (ramp of 40kPa/sec to maximum 1000kPa). Participants press a response button when discomfort/pain is experienced and PPT is taken as the mean of three repetitions of ascending stimuli applied either over the middle phalanx of the third finger of the non-dominant hand (digit PPT) or over the head of the fibula (knee PPT).

Dynamic Measures

Wind-up ratio (WUR). The weighted pinprick stimulator that produced a pricking sensation when calculating the MPT (above) is used to assess the WUR. The intensity from this single pinprick stimulus (VRS1) is compared to the perceived intensity following a 1/sec train of 10 repeated stimuli of the same intensity (VRS10), applied within a small area of 1cm². Participants are also asked if the stimulus feels the same, more intense (i.e. sensitization), or less intense (i.e. habituation). The WUR is calculated as VRS10 - VRS1.

Dynamic brush allodynia. Following 3 strokes with a calibrated brush (200-300 mN; SenseLab) for 1-2cm length over the skin, participants score the sensation between 0-10 (0=neutral; 10=unpleasant/painful).

Hyperalgesia Mapping

The area of hyperalgesia surrounding localised regions of pain or perceived sensory loss will be mapped by moving towards this in longitudinal and horizontal directions from distant sites. For subjects with potential peripheral sensory neuropathy, this will predominantly relate to moving proximally to distally in the limbs. For patients with prior surgery (e.g. PPSP patients), additional testing can be performed adjacent to scars.

Thermal hyperalgesia. The Somedic RollTemp® consists of two small hand-held rollers that are kept within a base unit between investigations and charged with thermal energy to their predetermined temperature levels of 25°C and 40°C. With a normal skin temperature of 32°C this corresponds to a

temperature difference of -7°C and $+8^{\circ}\text{C}$ respectively. Starting well away from the region of pain or scar, or commencing in the proximal limb, probes will be rolled at a rate of 1-2cm/sec, and the point where sensation is altered will be noted to non-invasively map areas of altered thermal sensitivity. If the sensation is unpleasant, cold and heat allodynia will be graded by the participant on a 0-10 verbal rating scale.

Mechanical stimuli. Mechanical sensitivity will be tested using hand-held calibrated von Frey hairs of increasing thickness (as described above for MDT). Hairs that deliver increasing pressure above the detection threshold will be sequentially applied until the stimulus is strong enough to produce mild discomfort, graded on a 0-10 verbal rating scale. Mechanical detection and discomfort thresholds will be determined with two series of ascending stimuli in a region outside the pain/wound area. From the distant site, a subthreshold hair (i.e. next hair down from that causing discomfort) will be applied along radial lines at intervals of 0.5cm, or from proximal to distal down the limb, until the first point where an altered sensation, increased sensitivity, discomfort, or a sore or sharp feeling is reported. The area of altered sensitivity will map the area of mechanical hyperalgesia. Within the hyperalgesic area, sequential von Frey hairs will be applied until discomfort or an altered sensation is reported. This value will be compared with the distant threshold to quantify the degree of hyperalgesia.

Brush allodynia. A calibrated brush will be moved towards the region of pain/scar and the degree of allodynia graded on a 0-10 verbal rating scale area, and the area of brush allodynia mapped.

Pain modulation

Cold pressor test. Participants immerse the hand up to the wrist with the palm down and fingers spread into a 5°C circulating water bath for 30 seconds, or prior to this if it became too uncomfortable or painful. The intensity of hand discomfort on removal is rated using a 0-10 verbal rating scale. This test will not be performed in subjects with a history of pain triggered by cold, or in those with significant cold allodynia of the hand identified in prior testing.

Conditioned pain modulation (CPM). Right knee PPT will be used as a variable test stimulus, and cold pressor test of the contralateral hand as the conditioning stimulus. This will ensure adequate distance between the test and conditioning stimuli, and also allow the right hand to be free for pressing the response button. Knee PPT will be measured at baseline (prior to hand immersion), during (at 15 secs), and after (50 and 90 secs following initial immersion) the cold pressor test.

Brain imaging

All scans will be carried out using a 3T MR scanner within the MRI Department at the Great Ormond Street Hospital NHS Trust. This provides resolution appropriate for assessing structural and functional changes in pain-related brain regions.

Test protocol: As described above, testing will encompass the following measures, as established in the Developmental Imaging & Biophysics section at the UCL GOS Institute of Child Health:

1. Grey and white matter integrity. These anatomical scans will be acquired first.
2. Resting state functional connectivity changes.

Sensory stimulation protocol: In a smaller sample of participants, the feasibility of using sensory stimuli within the scanner in our cohort will be assessed. These patients will have additional scans to assess stimulus-evoked functional activation and functional connectivity changes, lasting <30 mins; stimuli will include brush, von Frey hair and punctate probes.

10e. Safety considerations/Patient safety

All participating children are assessed by, and under the ongoing care of, the Chronic Pain Multidisciplinary Team. Questionnaires and QST are part of medical care, while neuroimaging is

being performed for assessment and is not a therapeutic intervention. Participants are informed that if the research results could influence the care of the child, this information will be shared with the clinical care team. They are also informed that brain scans will be used for research purposes only and will not be formally reviewed by a clinician.

10f. Data Monitoring

A randomly selected sample (10%) of database entries will be cross-checked for data entry errors by another member of the research team. There is no change to usual treatment and questionnaire and QST results will form part of the medical record.

10g. Prospective Collection of S-LANSS data (non-neuropathic pain)

As part of routine care, the S-LANSS is one of several patient-reported outcome measures (PROMs) completed by children and carers attending Chronic Pain Clinic (as outlined above). To facilitate comparison between children presenting with neuropathic or non-neuropathic pain, the S-LANSS data from children with chronic pain that is not clinically diagnosed as neuropathic will be prospectively collected. Following assessment of new patients at the Chronic Pain Clinic, or review of existing patients with non-neuropathic pain aged between 10 and 18 years, families will be identified by the clinical care team and given information related to the study, as described in Section 10a. Researchers will then approach the family and ask for parental consent and child assent for inclusion of clinical PROMs data in the current research proposal.

10h. Retrospective Case Note Review

As part of current routine care, patients from the Chronic Pain Service are referred for QST to a member of the clinical care team, who is also one of the current Investigators. The S-LANSS is completed with patients and QST performed using the same methodology as described in this proposal. Patients assessed prior to this proposal (currently n=30 children aged 10-18 years) include those with neuropathic pain and other causes of chronic pain, based on clinical history and evaluation at the Chronic Pain Clinic. We will make secondary use of this information previously collected in the course of usual clinical care. Data from the S-LANSS in patients both with and without neuropathic pain will be extracted and pseudonymised.

11. Statistical Considerations and Data Analysis

11a. Sample size and statistical power

We will recruit 74 children with neuropathic pain and 150 children with chronic pain of non-neuropathic origin. This sample size is based primarily on a power calculation to identify a suitable sample size for the primary objective, and is sufficient for the secondary objectives as well. Details are provided below (sample sizes and analysis are based on previous publications with similar patient groups):

Sensitivity & specificity of neuropathic pain screening tool. Sample size estimation was determined from a power calculation with values of sensitivity (0.74) and specificity (0.76) derived from an adult study, and based on a 0.33 prevalence of neuropathic pain in the Chronic Pain Outpatients service at GOSH. We used a conventional 95% confidence interval with 0.1 precision. Based on these values, the sensitivity analysis requires a minimum sample size of 224 children with chronic pain (including 74 with neuropathic pain). This sample size is also sufficient for the specificity analysis, which only requires 105 children (including 35 with neuropathic pain). Given an actual prevalence of 36%, the calculated sample size is a conservative estimate of the actual minimum sample size required.

Questionnaires. The most widely used questionnaire in our protocol (PedsQL, assessing quality of life) is used as a basis for sample size estimation. Normative values based on large cohorts are available for healthy children (mean \pm S.D. 82 ± 15.1) and for those with moderate (65.4 ± 19.6) and

major (56.4 ± 19.3) chronic conditions. Based on these means and standard deviations, we anticipate that a minimum of 18 children with neuropathic pain and 18 healthy children is required to achieve a minimum power of 80% to detect a difference between the two groups, with a target significance level of 0.05. Our neuropathic pain sample is more than sufficient and we already have control data available.

QST. QST descriptive analysis will be based on the somatosensory profile of sensory gain or loss (compared to existing normative data) in individual patients. A previous study with a similar sample size (42 children and adolescents with CRPS) used the same methodology and reported that the most common symptoms of children with CRPS are thermal and mechanical hyperalgesia. Similarly, the proportion of children having a sensory profile that matches clusters identified in adults with neuropathic pain will also be determined, to indicate potential parallels in children.

Neuroimaging. The primary aim is to assess feasibility of neuroimaging in this cohort, and to estimate recruitment rate. However, previous studies have found meaningful and statistically significant alterations in brain structure and function in paediatric cohorts ranging in size from 8 to 12 CRPS patients. Therefore, after taking into account potential data loss arising from technical issues such as motion artefact, an imaging recruitment rate of 16-27% within our sample of 74 would be sufficient for meaningful preliminary measures of brain alterations.

11b. Statistical methods

Sensitivity and specificity of the S-LANSS will be analysed in a group of children with chronic pain, of which 74 children have neuropathic pain, against the gold standard of clinical diagnosis.

Scores on patient- and parent-reported outcome measures in children with neuropathic pain will be compared with existing control data using independent samples t-tests or Mann-Whitney tests, if data are not normally distributed. Comparison with existing data in the chronic pain database on other causes of chronic pain in children is not the main focus of the current project and will be exploratory, to inform future research regarding likely effect size. Similarly, comparison with published control data will be descriptive.

A sensory profile will be described for each patient by comparison with existing healthy control data collected by the current investigators using the same methodology at 11 and 19 years of age, and with published control data in a large EU cohort of 176 children and adolescents. For each QST parameter, data will be z-transformed such that $Z\text{-score} = (X[\text{single patient}] - \text{Mean}[\text{controls}]) / \text{SD}[\text{controls}]$. This allows data to be compared, independent of the different units of measurement across the multiple sensory parameters. The proportion of patients having sensory changes for each QST parameter will be calculated. In addition, we will determine the proportion of patients with sensory profiles matching each of the three QST clusters identified in adults, and the proportion of profiles observed for each cause of neuropathic pain (e.g. persistent postsurgical pain; complex regional pain syndrome).

Neuroimaging: 1) Recruitment rate: proportion of patients consenting/assenting, and proportion completing neuroimaging at rest. 2) Feasibility of stimulation: ability of patients to lie still during stimulation/ability to obtain motion-free neuroimaging data; acceptability by patients/parents. If a pain score greater than 8/10 is reported in response to any stimulus, scanning during stimulation will cease. 3) Preliminary analysis of neuroimaging parameters at rest: Paired t-tests comparing grey and white matter integrity between patients and existing control data. Paired t-tests comparing correlation values (representing functional connectivity between key brain regions) between patients and existing control data. 4) Analysis of neuroimaging parameters during stimulation: Paired t-tests comparing activation following stimulation of the affected and unaffected body areas within patients

(effect of body area). All analysis of neuroimaging data will be corrected for multiple comparisons using the false discovery rate.

12. Ethical considerations

The study will be conducted in full conformance with principles of the Declaration of Helsinki, Good Clinical Practice (GCP), and within the laws and regulations of the UK. All investigators will receive regular training in GCP and in safeguarding children. The protocol has been reviewed and approved by the NHS Health Research Authority West Midlands – Black Country Research Ethics Committee on 23rd August 2017 (REC reference 17/WM/0306; IRAS Project ID 226141).